

IN THE CLAIMS:

Please amend the claims as follows:

1. (Currently amended) A method of detecting a target nucleic acid in a sample suspected of comprising the target nucleic acid, the method comprising:

providing a hybridization complex comprising (a) a capture probe that is attached to a solid surface, (b) the target nucleic acid hybridized to the capture probe, and (c) at least one nanoparticle attached to the target nucleic acid, wherein the nanoparticle is spherical, has a diameter of less than 1000 nanometers, and comprises a material known to absorb light at one or more particular wavelength, and wherein the capture probe comprises at least one oligonucleotide that is complementary in whole or in part to the target nucleic acid, and wherein providing the hybridization complex is performed under conditions wherein the capture probe selectively hybridizes to the target nucleic acid;

removing unattached nanoparticle and unhybridized nucleic acid from the solid surface;

exposing the solid surface to light at a wavelength absorbed by the nanoparticle and not by the solid surface;

detecting a temperature of the solid surface in the presence of said complex;

providing a background temperature of the solid surface in the absence of said complex; and

comparing the temperature of the solid surface detected in the presence of said complex with the background temperature, whereby detection of an increased temperature in the presence of said complex relative to the background temperature indicates the presence or amount of target nucleic acid in the sample.

2. (Previously presented) The method according to claim 1, comprising:

hybridizing the target nucleic acid to at least one capture probe to form a first hybridization complex, wherein the capture probe is attached to a solid surface at an attachment location;

attaching a detection probe to the first hybridization complex to form a second hybridization complex, wherein the detection probe comprises the nanoparticle;

exposing the solid surface to light at a wavelength absorbed by the nanoparticle; and

detecting the temperature of the solid surface at the attachment location of the capture probe, wherein an increase in temperature at the attachment location as compared to the background temperature of the solid surface indicates hybridization of the target nucleic acid.

3. (Previously presented) The method of Claim 1, wherein the target nucleic acid comprises RNA.

4. (Previously presented) The method of Claim 1, wherein the target nucleic acid comprises cDNA.

5. (Original) The method according to Claim 1, wherein the solid surface comprises indium tin oxide.

6. (Previously presented) The method according to Claim 1, wherein the sample is a biological sample.

7. (Previously presented) The method according to Claim 2, wherein the nanoparticle comprises one or more of the group consisting of a metal and a metal oxide.

8. (Previously presented) The method according to Claim 7, wherein the nanoparticle comprises a metal comprising one or more of the group consisting of gold, silver, and platinum.

9. (Original) The method according to Claim 1, wherein the nanoparticle comprises gold.

10. (Original) The method according to Claim 1, wherein the nanoparticle is a nanoshell.

11. (Previously presented) The method according to Claim 1, wherein the nanoparticle has a diameter from about 10 to about 20 nanometers.

12. (Original) The method according to Claim 1, wherein the nanoparticle exhibits surface plasmon resonance, and wherein the solid surface is exposed to light at a wavelength that matches the surface plasmon resonance of the nanoparticle.

13. (Original) The method according to Claim 1, wherein the light is generated by a laser.

14. (Original) The method according to Claim 2, wherein the detection probe further comprises an oligonucleotide attached to the nanoparticle.

15. (Previously presented) The method according to Claim 14, wherein the capture probe is complementary to a first target domain of the target nucleic acid, and the detection probe oligonucleotide is complementary to a second target domain of the target nucleic acid.

16. (Previously presented) The method according to Claim 2, wherein the detection probe comprises the nanoparticle attached to one partner of a ligand-binding pair, and the target nucleic acid comprises the other partner of a ligand-binding pair.

17. (Previously presented) The method according to Claim 16, wherein one partner of the ligand-binding pair is streptavidin, and the other partner of the ligand-binding pair is biotin.

18. (Previously presented) The method according to Claim 16, wherein the target nucleic acid comprises biotin.

19. (Previously presented) The method according to claim 18, wherein the biotin has been incorporated into the target nucleic acid during nucleic acid amplification.

20. (Original) The method according to Claim 18, wherein the detection probe comprises a nanoparticle attached to streptavidin.

21. (Original) The method according to Claim 1, wherein a plurality of different capture probes are attached to the solid surface in an array, and the location of each capture probe comprises an array element.

22. (Original) The method according to Claim 21, wherein each array element is exposed to light separately.

23. (Original) The method according to Claim 21, wherein the entire plurality of capture probes is exposed to light simultaneously.

24. (Previously presented) The method according to Claim 1, wherein the light is provided by a light source selected from the group consisting of a tungsten halogen light source, a xenon arc lamp and a laser.

25. (Original) The method according to Claim 1, where in the exposing is by rastering.

26. (Previously presented) The method according to Claim 1, wherein the target nucleic acid is selected from the group consisting of an mRNA and a cDNA.

27. (Previously presented) The method according to Claim 1, wherein the capture probe comprises an oligonucleotide from a gene of interest.

28. (Previously presented) The method according to Claim 1, wherein the capture probe comprises a mutation to be detected.

29. (Previously presented) The method according to Claim 1, wherein the target nucleic acid comprises or is suspected to comprise a mutation to be detected.

30. (Original) The method according to Claim 1, wherein the nanoparticle comprises silver and the solid surface is exposed to light at a wavelength ranging from about 420-460 nm.

31. (Original) The method according to Claim 1, wherein the nanoparticle comprises gold and the solid surface is exposed to light at a wavelength of about 532 nm.

32. (Original) The method according to Claim 1, wherein the detecting step is carried out by a thermocouple attached to a side of the solid surface upon which capture probes are not attached.

33. (Original) The method according to Claim 1, wherein the detecting step is carried out by infrared thermography.

34. (Original) The method according to Claim 1, wherein the detecting step is carried out by Fourier Transform infrared thermography.

35. (Original) The method according to Claim 1, wherein the detecting step comprises capturing a thermal image by means of an infrared camera.

36. (Original) The method according to Claim 1, wherein the detecting step is carried out by a charge coupled device.

37. (Canceled)

38. (Previously presented) The method according to claim 1, where the nanoparticle is attached to the target nucleic acid by one of the group consisting of a binding pair and complementary nucleic acids.

39. (Previously presented) The method according to claim 1, where the nanoparticle is attached to the target nucleic acid by one of the group consisting of primer extension and ligation of a nanoparticle-labeled nucleic acid.

40. (Previously presented) The method of claim 1, wherein the hybridization complex comprises a detection probe, wherein the detection probe comprises the nanoparticle.

41. (Previously presented) The method of claim 40, wherein the detection probe is attached to the target nucleic acid before, during, or after the target nucleic acid hybridizes to the capture probe.

42. (Previously presented) The method of claim 40, comprising the sequential steps of hybridizing the target nucleic acid to the capture probe to form a hybrid; and then reacting the hybrid with a detection probe.

43. (Previously presented) The method of claim 1, wherein the hybridization complex is present at a concentration of at least 10 fM.

44. (Currently amended) A method of detecting a target nucleic acid in a sample suspected of comprising the target nucleic acid, the method comprising:

providing an at least 10 fM concentration of a hybridization complex comprising (a) a capture probe that is attached to a solid surface and (b) a target nucleic acid hybridized to the capture probe, and (c) at least one nanoparticle attached to the target nucleic acid, wherein the nanoparticle comprises a metal or metal oxide that exhibits surface plasmon resonance, is spherical, and has a diameter of less than 1000 nanometers, the solid surface is a material that is different than the nanoparticle, and wherein the capture probe comprises at least one oligonucleotide that is complementary in whole or in part to the target nucleic acid and wherein providing the hybridization complex is performed under conditions wherein the capture probe selectively hybridizes to the target nucleic acid;

removing unattached nanoparticle and unhybridized nucleic acid from the solid surface;

exposing the solid surface to light at a wavelength that matches the surface plasmon resonance of the nanoparticle and is not absorbed by the solid surface; and

detecting a temperature of the solid surface in the presence of said complex;

providing a background temperature of the solid surface in the absence of said complex; and

comparing the temperature of the solid surface detected in the presence of said complex with the background temperature, whereby detection of an increased temperature of the solid surface in the presence of said complex relative to the background temperature indicates the presence or amount of target nucleic acid in the sample.

45. (Currently amended) A method of detecting a target nucleic acid in a sample suspected of comprising the target nucleic acid, the method comprising:

(a) providing an at least 10 fM concentration of a hybridization complex comprising:

(ia) a capture probe that is attached to a solid surface-selected from glass and indium tin oxide, the capture probe comprising an oligonucleotide of 5 to 50 nucleotides;

(bij) a target nucleic acid hybridized to the capture probe, wherein the target nucleic acid comprises an oligonucleotide of between 10 and 300 nucleotides; and

(eijj) at least one spherical gold nanoparticle having a diameter of between 5 and 200 nm attached to the target nucleic acid sequence,; wherein the oligonucleotide of (i) is complementary to the target nucleic acid;

wherein the target nucleic acid is incubated with the capture probe at a sodium ion concentration of less than 1.0 M, a pH of between 7.0 and 8.3, and at a temperature of at least 30°C;

wherein providing the hybridization complex is performed under conditions such that the capture probe selectively hybridizes to the target nucleic acid; and

wherein any unhybridized nucleic acid and unattached nanoparticle is removed; and

(b) exposing the solid surface to light at a wavelength of between about 510 nm and about 560 nm; and

(c) detecting a temperature of the solid surface in the presence of said complex;

(d) providing a background temperature of the solid surface in the absence of said complex; and

(e) comparing the temperature of the solid surface detected in the presence of said complex with the background temperature, whereby detection of an increased temperature of the solid surface in the presence of said complex relative to the background temperature indicates the presence or amount of target nucleic acid in the sample;

wherein providing a hybridization complex further comprises:



~~providing a capture probe attached to a solid surface selected from glass and indium tin oxide (ITO), the capture probe comprising an oligonucleotide of 5 to 50 nucleotides that is complementary to the target nucleic acid;~~

~~— providing a target nucleic acid comprising an oligonucleotide of between 10 and 300 nucleotides;~~

~~incubating the target nucleic acid with the capture probe at a sodium ion concentration of less than 1.0 M, a pH of between 7.0 and 8.3, and at a temperature of at least 30°C; and~~

~~removing any unhybridized nucleic acid.~~

46. (Previously presented) The method of claim 1, wherein the sample comprises a heterogeneous nucleic acid mixture and providing the hybridization complex comprises selecting hybridization conditions wherein the capture probe hybridizes to the target nucleic acid to the exclusion of other nucleic acids present in the sample.